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FORM PTO-1390 (Modified) (REV 11-98) U.S. DEPARTMENT OF COMMERCE PATENT AND TRADEMARK OFFICE

TRANSMITTAL LETTER TO THE UNITED STATES DESIGNATED/ELECTED OFFICE (DO/EO/US) CONCERNING A FILING UNDER 35 U.S.C. 371

ATTORNEY'S DOCKET NUMBER: 27611/35364

U S APPLICATION NO (IF KNOWN, SEE 37 CFR): 09/622816

INTERNATIONAL APPLICATION NO	INTERNATIONAL FILING DATE	PRIORITY DATE CLAIMED
PCT/US99/03805	22 February 1999	24 February 1998

TITLE OF INVENTION
LIPID EMULSIONS IN THE TREATMENT OF SYSTEMIC POISONING

RECEIVED
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TECH CENTER 1600/2900

APPLICANT(S) FOR DO/EO/US
WEINBERG, Guy and HERTZ, Paul

Applicant herewith submits to the United States Designated/Elected Office (DO/EO/US) the following items and other information:

1. ☒ This is a **FIRST** submission of items concerning a filing under 35 U.S.C. 371
2. ☐ This is a **SECOND** or **SUBSEQUENT** submission of items concerning a filing under 35 U.S.C. 371.
3. ☐ This is an express request to begin national examination procedures (35 U.S C. 371(f)) at any time rather than delay examination until the expiration of the applicable time limit set in 35 U.S.C. 371(b) and PCT Articles 22 and 39(1).
4. ☒ A proper Demand for International Preliminary Examination was made by the 19th month from the earliest claimed priority date.
5. ☒ A copy of the International Application as filed (35 U.S.C. 371 (c) (2))
   a. ☒ is transmitted herewith (required only if not transmitted by the International Bureau).
   b. ☐ has been transmitted by the International Bureau.
   c. ☐ is not required, as the application was filed in the United States Receiving Office (RO/US).
6. ☐ A translation of the International Application into English (35 U.S.C. 371(c)(2)).
7. ☒ A copy of the International Search Report (PCT/ISA/210).
8. ☒ Amendments to the claims of the International Application under PCT Article 19 (35 U.S.C. 371 (c)(3))
   a. ☐ are transmitted herewith (required only if not transmitted by the International Bureau).
   b. ☐ have been transmitted by the International Bureau.
   c. ☐ have not been made; however, the time limit for making such amendments has NOT expired.
   d. ☒ have not been made and will not be made.
9. ☐ A translation of the amendments to the claims under PCT Article 19 (35 U.S.C. 371(c)(3)).
10. ☐ An oath or declaration of the inventor(s) (35 U S C 371 (c)(4)).
11. ☐ A copy of the International Preliminary Examination Report (PCT/IPEA/409).
12. ☐ A translation of the annexes to the International Preliminary Examination Report under PCT Article 36 (35 U.S.C. 371 (c)(5)).

Items 13 to 20 below concern document(s) or information included:

13. ☐ An Information Disclosure Statement under 37 CFR 1.97 and 1.98.
14. ☐ An assignment document for recording A separate cover sheet in compliance with 37 CFR 3.28 and 3.31 is included.
15. ☐ A **FIRST** preliminary amendment.
16. ☐ A **SECOND** or **SUBSEQUENT** preliminary amendment.
17. ☐ A substitute specification.
18. ☐ A change of power of attorney and/or address letter
19. ☒ Certificate of Mailing by Express Mail
20. ☒ Other items or information:

Small Entity Claim Form - Non-Profit Organization

PCTUS1/REV03

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U.S. APPLICATION NO (IF KNOWN, S[illegible] 7 CFR	INTERNATIONAL APPLICATION NO	ATTORNEY'S DOCKET NUMBER
09/622816	PCT/US99/03805	27611/35364

21. The following fees are submitted..

BASIC NATIONAL FEE (37 CFR 1.492 (a) (1) - (5)) :		CALCULATIONS PTO USE ONLY
☐ Neither international preliminary examination fee (37 CFR 1.482) nor international search fee (37 CFR 1 445(a)(2) paid to USPTO and International Search Report not prepared by the EPO or JPO	$970.00	
☐ International preliminary examination fee (37 CFR 1.482) not paid to USPTO but Internation Search Report prepared by the EPO or JPO	$840.00	
☐ International preliminary examination fee (37 CFR 1 482) not paid to USPTO but international search fee (37 CFR 1 445(a)(2)) paid to USPTO	$690.00	
☒ International preliminary examination fee paid to USPTO (37 CFR 1.482) but all claims did not satisfy provisions of PCT Article 33(1)-(4)	$670.00	
☐ International preliminary examination fee paid to USPTO (37 CFR 1.482) and all claims satisfied provisions of PCT Article 33(1)-(4)	$96.00	

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ENTER APPROPRIATE BASIC FEE AMOUNT =		$670.00
Surcharge of **$130.00** for furnishing the oath or declaration later than ☐ 20 ☒ 30 months from the earliest claimed priority date (37 CFR 1.492 (e)).		$130.00

CLAIMS	NUMBER FILED	NUMBER EXTRA	RATE		
Total claims	47 - 20 =	27	x $18.00	$486.00	
Independent claims	4 - 3 =	1	x $78.00	$78.00	
Multiple Dependent Claims **(check if applicable)**.			☒	$260.00	
	TOTAL OF ABOVE CALCULATIONS		=	$1,624.00	
Reduction of 1/2 for filing by small entity, if applicable. Verified Small Entity Statement must also be filed (Note 37 CFR 1.9, 1.27, 1.28) **(check if applicable)**.			☒	$812.00	
	SUBTOTAL		=	$812.00	
Processing fee of **$130.00** for furnishing the English translation later than ☐ 20 ☐ 30 months from the earliest claimed priority date (37 CFR 1.492 (f)).			+	$0.00	
	TOTAL NATIONAL FEE		=	$812.00	
Fee for recording the enclosed assignment (37 CFR 1.21(h)). The assignment must be accompanied by an appropriate cover sheet (37 CFR 3.28, 3.31) **(check if applicable).**			☐	$0.00	
	TOTAL FEES ENCLOSED		=	$812.00	
				Amount to be: refunded	$
				charged	$

☒ A check in the amount of **$812.00** to cover the above fees is enclosed.

☐ Please charge my Deposit Account No. in the amount of to cover the above fees.
A duplicate copy of this sheet is enclosed.

☒ The Commissioner is hereby authorized to charge any fees which may be required, or credit any overpayment to Deposit Account No. **13-2855** A duplicate copy of this sheet is enclosed.

NOTE: Where an appropriate time limit under 37 CFR 1.494 or 1.495 has not been met, a petition to revive (37 CFR 1.137(a) or (b)) must be filed and granted to restore the application to pending status.

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36,107
REGISTRATION NUMBER

August 23, 2000
DATE

LIPID EMULSIONS IN THE TREATMENT OF SYSTEMIC POISONING

Field of the Invention

This invention relates to lipid emulsion compositions and to methods of reducing the bioavailability and toxicity of poisons and other noxious agents present in the circulation by the intravenous infusion of a lipid emulsion composition.

Background of the Invention

An emulsion is a collective of lipid or oil microparticles dispersed in water usually by the action of an emulsifying agent. Historically, emulsions have been widely used in the cosmetic and drug industries in connection with creams, depilatories, antiperspirants, deodorants, antiseptics and the like. Emulsion systems which include sugars, amino acids, vitamins, and electrolytes have also been utilized as a means of providing intravenous nutrition in order to maintain a patient's life when oral or nasal feeding is impossible or insufficient (see U.S. Patent No. 5,674,527 to Inoue et al.). Another common use of emulsions is in parenteral drug delivery systems (see U.S. Patent No. 3,538,216 to Polin et al.). These drug delivery systems feature hydrophobic medicinals suspended in an emulsion to be delivered to the patient in a sustained release manner. The lipid emulsion in this system consists of a thixotropic agent, a gelatinous-oil composition containing an ion-exchange agent, and water.

Also, edible but non-digestible emulsions have been used as traps for toxins present in the gastrointestinal (GI) tract as described in U.S. Patent No. 4,183,918 to Asher et al. In this trap system, the emulsion is fed to a symptomatic patient wherein the toxins are removed by the action of the absorbent-containing emulsion passing through the GI tract. Key features of this system include the use of non-digestible oils as the exterior phase of the emulsion and the use of a reactant or adsorbent in the interior aqueous phase of the emulsion. Examples of exterior phase oils used in this system include highly refined hydrocarbon oils, mineral oils, and silicone oils, while preferred interior phase reactants and adsorbents include silica gel and carbon.

Other means of detoxifying the body include the delivery of liposomes containing active reagents to a patient. For example, an aqueous solution of the

chelating agent EDTA was encapsulated by liposomes (synthetic membrane vesicles) and given to a patient undergoing chemotherapy in order to remove the radioactive metal plutonium from the patient's body. (Rahman et al., Science (1973) **180**:300). Liposomes, in most cases, act by rupturing their membranes to release their inner contents. As such, liposomes have also been used to deliver drugs in a controlled-release manner as described in U.S. Patent No. 4,837,028 to Allen. However, liposomes are not readily permeable to extraneous toxic agents present in the body.

Despite the foregoing, a need remains for materials and methods to effectively decrease the bioavailability and toxic substances in the bloodstream, especially those lipophilic or amphilic agents such as antidepressants, anesthetics, alcohol, or others which require immediate intervention when present in dangerous amounts.

Summary of the Invention

The invention is directed to lipid emulsion compositions and methods for reducing the bioavailability and toxicity of poisonous agents in the circulation by the intravenous infusion of a lipid emulsion. Preferred lipid emulsion compositions comprise an oil, an emulsifier, a tonicity modifier, and water. In a preferred method of the invention, a patient having toxic levels of a drug or other toxic substances is intravenously infused with a composition of the lipid emulsion wherein the toxic substance permeates the emulsion and is redistributed according to its lipid:aqueous partition coefficient into the non-aqueous (lipid) phase of the emulsion, thereby decreasing the bioavailability of the toxic substance. Such lipid sinks have wide applicability to the treatment of toxicity associated with lipophilic and amphiphilic substances. In a preferred embodiment, the invention is directed to the treatment of toxicity due to lipophilic and amphiphilic substances.

In another preferred embodiment, the invention is useful in the treatment of cardiotoxicity, including those instances of cardiac arrest due to unknown toxic agents and, in particular, when the toxic agents are lipophilic and amphiphilic substances.

Yet another embodiment of the present invention comprises materials and methods for treating toxicity associated with anesthetic agents, including but not limited to, bupivacaine, lidocaine, and other anesthetic agents.

A preferred lipid emulsion composition comprises about 20 percent by weight soybean oil, about 2 weight percent glycerin, about 1 weight percent egg yolk phospholipid, and about 80 weight percent water; however, the composition can vary depending upon the nature and lipid partition coefficient of the toxic substance in the bloodstream.

In a preferred embodiment, a 20 percent by weight solution of the emulsion in water is infused intravenously at an initial rate of about 7.5 milliliters per kilogram for a time period of about 30 seconds followed by a steady-state rate of about 3 milliliters per kilogram per minute for a time period of about 2 minutes.

Other lipid-emulsions according to the present invention include emulsions comprising one or more of the following substances: glycerophospholipids such as phosphatidylcholine; cholesterol, stearylamine; phosphatidylserine; phosphotidylglycerol and other lipids. Also included within the scope of the invention are microemulsions which include oil, water, and an amphiphile system that is macroscopically mono-phasic, optically isotropic, thermodynamically stable and characterized by ultra-low interfacial tension values.

The invention is also directed to a device for the convenient administration of the emulsions of the present invention to a patient. The device is also useful for administering other therapeutic substances by way of a regimen comprising the administration of a bolus of the agent and the subsequent infusion of the agent over a period of time.

Brief Description of the Drawings

Figure 1 is a graph depicting the probit analysis of the mortality fraction versus the bupivacaine dose for animals treated according to protocol 2 (lipid resuscitation) for intravenous infusions of either saline or a lipid emulsion composition.

Figure 2 depicts a device for administering to a patient the emulsions of the present invention.

Detailed Description

The present invention relates to lipid emulsion compositions and to methods and apparatus for reducing the bioavailability and toxicity of poisonous or noxious agents present in the circulation, by intravenous administration of a lipid emulsion. A preferred method for the treatment of systemic toxicity includes making a patient in need of such therapy, rapidly lipemic by the intravenous infusion of an initial large bolus dose of an emulsion followed by a slower steady-state rate infusion of the emulsion. Although the rate of infusion can vary with respect to the particular emulsion utilized with the toxic agent involved and with the particular patient, by way of example, an initial rate of the infusion may be in the range of about 0.5 ml/kg/min to about 10 ml/kg/min for a time period of about 0.5 min followed by a steady-state rate in the range of about 0.1 ml/kg/min to about 3 ml/kg/min for a time period of about 2 minutes.

The lipid emulsion composition used to detoxify the blood comprises an oil, an emulsifier, a tonicity modifier, and water. Additional ingredients can include a surfactant, a co-solvent, a bacteriostat, a preservative, an active ingredient, and an adsorbent.

Preferably the oil in the emulsion composition is one or more oils selected from the group consisting of monoglycerides, diglycerides, triglycerides, and mixtures thereof. More preferably, the oil is a naturally occurring plant oil selected from the group consisting of soybean oil, cottonseed oil, safflower oil, corn oil, coconut oil, sesame oil, peanut oil, olive oil, and mixtures thereof. Most preferably the oil is soybean oil. In addition, the oil can be an animal oil or a fish oil such as cod liver oil. The oil can also can be a mineral oil or a chemically-synthesized oil such as 2-linoleoyl-1,3-dioctanoyl glycerol. Semisynthetic mono-, di- or triglycerides may also be used and include rac-glyceryl-1-monopalmitic, acyl glyceryl-1-monoolein, 1, 2-dipalmitic, 1, 3-dipalmitic, trimyristin, tripalmitin, tristearin, triolein, trilaiden and the like.

The emulsifier in the lipid emulsion composition preferably is a naturally-occurring phospholipid. Preferred phospholipids can be derived from egg or soy sources. Exemplary phospholipids include but are not limited to, egg yolk

phospholipids, hydrogenated egg yolk phospholipids, soybean phospholipids, hydrogenated soybean phospholipids, and mixtures thereof. Preferably, the phospholipid is egg yolk phospholipid. The emulsifier also can be a synthetic lecithin such as dihexanoyl-L-∝-lecithin. Among the other emulsifiers useful in the practice of the present invention are other glycerophospholipids such phosphatidylcholine lipids such as cholesterol, stearylamine, phosphatidylserine, phosphatidylglycerol and other lipids.

The tonicity modifier preferably is a member of the group consisting of glycerin, sorbital, polyoxyethylated hydrocarbons, and C_6-C_{20} saturated or unsaturated aliphatic acids. The optional co-solvent preferably is an alcohol such as isopropanol or benzyl alcohol or the like. The bacteriostat or preservative can be any of those commercially available which are non-toxic. The active ingredient can be a desired drug or reactant which can render the toxic agent non-toxic or which may act to counter the physiological effects of the toxic agent, while the adsorbent can be, for example, charcoal, silica gel, or the like.

In formulating the emulsion, the oil is preferably present in an amount in the range of about 10 to about 30 percent by weight of the composition. The surfactant in the emulsion composition is present in an amount in the range of about 1 to about 5 percent by weight of the composition. Water is present in the emulsion composition in an amount in the range of about 70 to about 90 percent by weight.

A preferred lipid emulsion composition comprises about 20 weight percent soybean oil, about 2 weight percent glycerin, about 1 weight percent egg yolk phospholipid, and about 80 weight percent water.

In the following examples, the commercially available (Healthcare, Deerfield, IL) lipid emulsion composition, Intralipid® was used. Intralipid® (Baxter) was introduced into the U.S. marketplace in 1975 for intravenous use. Intralipid® contains 10% w/v soybean oil as a source of polyunsaturated fatty acids, and 1.2% w/v of purified egg phospholipids which act as an emulsifying agent. The remainder of the composition is water added to achieve final lipid concentration in the range of about 10% w/v to about 30%w/v as is desired. Glycerol is added to make the lipid emulsion

isotonic, with about 2.25% w/v present in Intralipid®. The pH range of the Intralipid® emulsion is from about 5.5 to 8.

The lipid emulsion can be prepared by any convenient means, such as sonication and the like. The components of the emulsion can be mixed or premixed in any order prior to the sonication process. The emulsion preferably comprises particles in the range of about 0.25 microns to about 0.75 microns in diameter.

While the invention is exemplified by way of reducing or eliminating the toxic effects of the anaesthetic, bupivacaine, it is readily apparent to one of skill in the art that lipid emulsions may also be used to treat toxicity associated with other lipophilic or amphiphilic agents including tricyclic antidepressants (e.g., amitryptiline), adriamycin, organic solvents, other anesthetic agents such as tetracaine, etidocaine and alcohol.

The methods and compositions of the present invention are applicable to several clinical scenarios in addition to treatment of acute toxicity such as is exemplified below. For example, in the situation where a patient will be receiving a known amount of toxin (e.g., a lipophilic chemotherapeutic agent such as adriamycin), an emulsion according to the invention may be administered to the patient to reduce toxicity of the agent thereby increasing its safe dose.

In another scenario, when an acutely ill patient presents with apparent toxicity or a possible overdose of a known or unknown drug, e.g., presenting with cardiac arrhythmias in a young, otherwise healthy person, or a person with a history of depression being treated with tricyclic antidepressants, the patient may be treated with an emulsion according to the present invention.

Other exemplary lipophilic toxic agents which may be sequestered using the emulsions of the present invention include gasoline, inhaled propellants, N, N-diethyl-m-toluamide (DEET).

The amount of toxin might be known precisely, or entirely unknown. In the latter case, the patient's clinical status (mildly or severely ill) will guide treatment. The length of treatment following an initial dose will be determined by clinical response against a predetermined maximum safe dose for a patient's weight, which is readily determined by routine methods. The spent emulsion will be

metabolized slowly (over hours) probably by lipoprotein lipase which releases the fatty acids from the triglycerides. The toxin is then released from the emulsion droplets, but this slow release allows the patient's normal metabolism to chemically modify, excrete, or otherwise detoxify the toxin. The emulsion can be delivered via any peripheral or central vein.

The invention is described in more detail below by way of non-limiting examples. Example 1 describes the pretreatment of animals with a lipid emulsion and the effect of shifting the dose-response to bupivacaine induced asystole. Example 2 demonstrates the resuscitation of an animal from a toxic dose of bupivacaine by use of an intravenously infused lipid emulsion.

Example 1

Pretreatment with a Lipid Emulsion Composition Shifts the Dose-Response to Bupivacaine Induced Asystole in Rats

Studies were undertaken to assess the ability of a lipid emulsion to shift the dose-response to drug-induced asystole (heart stoppage) in rats. Pretreatment with a lipid emulsion increased the dose of bupivacaine (a local anaesthetic) required to induce asystole. Racemic bupivacaine hydrochloride was purchased from Sigma (St. Louis, MO) while tritiated bupivacaine was purchased from Moravek Biochemicals (Brea, CA). Intralipid® was purchased from Baxter Healthcare (Deerfield, IL). Male Sprague-Dawley rats weighing between about 250 grams to about 370 grams were used in all experiments.

Animals were first anesthetized in a bell jar with isoflurane to allow intubation, then mechanically ventilated with about 1.75% isoflurane in about 100% oxygen using a Harvard rodent ventilatory model 680 in conjunction with a tidal volume of 3 ml and a starting rate of about 40 breaths per minute. Catheters were inserted into the right internal jugular vein, the right carotid artery, and the right internal iliac vein. Electrocardiogram (ECG) was monitored via three subcutaneous needle electrodes in each rat. Arterial blood gas measurements were made after the induction of general anesthesia and again just prior to infusions to confirm a pCO_2 in

the range of about 30 to about 35 mm Hg and a pH in the range of about 7.35 to about 7.45 units.

All animals were allowed to stabilize for about 15 minutes while arterial blood pressure and ECG were monitored. There were six animals in each group. Control animals (group 1) received saline intravenously as pretreatment while test animals (groups 2-4) were pretreated intravenously with the lipid emulsion composition Intralipid® at concentrations of either about 10% by weight (group 2), about 20% by weight (group 3) , or about 30% by weight (group 4) in saline. All pretreatments were infused at a rate of about 3 ml/kg/min for 5 minutes via the internal jugular vein.

Immediately following pretreatment, all animals received an infusion of about 0.75% bupivacaine via the internal iliac catheter at a rate of 10 mg/kg/min to an end point of about ten seconds of asystole. Blood was then drawn from the aorta into a heparinized syringe for plasma bupivacaine determinations. The cumulative lethal dose of bupivacaine was calculated in mg/kg for all animals.

Plasma bupivacaine concentrations were determined by high performance liquid chromatograph (HPLC) after the samples had been extracted with hexane. The method of hexane extraction was validated with bupivacaine spiked samples and provided greater than about 95% recovery of bupivacaine from both normal and lipemic plasma. Thus, plasma bupivacaine concentrations reflected total bupivacaine content in both the aqueous and lipid phases of the specimen. The compounds were separated using a C18 column, 5 micrometer, 150 by 3.9 mm internal diameter (Symmetry, Waters Associates, Milford, MA) using the anaesthetic mepivacaine as an internal standard. The mobile phase consisted of about 25% acetonitrile in about 25 mM phosphate buffer adjusted to about pH 3.0. The flow rate was about 1 ml/min with a constant column temperature of about 30°C. The retention time for the mepivacaine internal standard was about 1.8 min. and about 4.0 min for bupivacaine during a 6 min. long run. The drugs were detected at a wavelength of about 215 nm.

The bupivacaine lipid:aqueous partition coefficient was determined for a mixture of Intralipid® and rat plasma. Blood obtained from rats by direct heart

puncture under halothane anesthesia was centrifuged and the plasma was separated. Equal volumes of about 30% Intralipid® and plasma (approximately 2 ml each) were combined and vortexed. Approximately 1.0 μCi of tritiated bupivacaine (specific activity 0.81 Ci/mole) was added to the mixture to a final bupivacaine concentration of about 93 μg/ml. This mixture was vortexed again then separated into aliquots of about 1 ml. These aliquots were allowed to sit undisturbed for about one hour at about 38°C, then centrifuged at about 10,000 g for about 10 minutes. High speed centrifugation separated each of these mixtures into a clear aqueous phase (about 0.85 ml) under a lipid phase (about 0.15 ml). The latter comprised a clear layer beneath a very thin white cap. The cap was removed then redissolved in saline to a total volume of about 1 ml. Aliquots of this solution and the aqueous plasma phase were then analyzed for tritiated bupivacaine content by liquid scintillation counting. The bupivacaine lipid:aqueous partition coefficient was given by the ratio of bupivacaine in the combined lipid phase (following correction for saline dilution) to the bupivacaine in the aqueous phase. This experiment was performed in triplicate.

Bupivacaine dose and plasma concentrations were analyzed by Kruskal-Wallis one way analysis of variance on ranks. Post hoc testing of both data sets was performed by Student-Newman-Keul's method for multiple comparisons (SigmaStat, Jandel Scientific /San Rafael, California). Cumulative bupivacaine dose data were nonparametric and median values were compared by differences of ranks. Plasma bupivacaine concentration data were parametric and differences in mean values were evaluated. Probit analysis (CalcuSyn, Biosoft/Cambridge, England) was used to compare bupivacaine LD_{50} values in the saline and lipid portions. The difference in survival of the two groups at 15mg/kg bupivacaine was further evaluated using a z test of proportions. Statistical significance in all experiments was taken as p less than or equal to 0.05.

The results indicate that the lethal bupivacaine dose among all animals ranged from about 12.7 mg/kg in an animal receiving saline pretreatment to about 111 mg/kg in an animal receiving an emulsion composition containing about 30% Intralipid®. Median bupivacaine lethal doses were as follows: (mg/kg; 25th percentile-75th percentile): Group 1 (saline) 17.8, 13.2-20.3; Group 2 (10% Intralipid®) 27.6,

22.2-31.7; Group 3 (20% Intralipid®) 49.8, 41.2-57.8; Group 4 (30% Intralipid®) 82.0, 71-3-101. Statistical significance for differences in median lethal bupivacaine does was achieved between all groups (p less than 0.001).

The mean plasma bupivacaine concentrations at the time of asystole for protocol 1 were (mcg/ml +/- standard error of the mean): group 1, 93.3+/-7.6; group 2, 115+/-15; group 3, 177+/-31; and group 4, 212+/-45. Statistical significance was achieved for the difference in mean concentrations between groups 1 and 4.

Probit analysis of the data from protocol provided the following bupivacaine LD_{50} values for the two treatment groups [lower and upper 95% confidence intervals (mg/kg): saline, 12.5, 11.8-13-4; lipid, 18.5, 17.8-19.3]. A z test of proportions at 15 mg/kg bupivacaine showed significance in the difference in survival between the two groups at this dose (p less than 0.004).

The lipid:aqueous ratio of bupivacaine concentrations (+/- standard error) was 11.9+/-1.77. When equal volumes of a solution of about 30% by weight Intralipid© and plasma were combined, the actual lipid volume was about 15% of total, and the percent of total bupivacaine dissolved in the lipid phase of this mixture (+/- standard error) was about 75.3%+/-1.32%

Example 2

Resuscitation from a Toxic Dose of Bupivacaine with a Lipid Emulsion Composition.

Experiments were performed to evaluate the ability of a lipid emulsion to resuscitate an animal from a toxic dose of bupivacaine. All animals were anaesthetized, instrumented and stabilized at about 1.75% isoflurane as described in Experiment 1, and arterial blood pressure and ECG were continuously monitored. Each rat received an intravenous dose of bupivacaine (see below for doses) for more than about 10 seconds by Harvard infusion pump, via the iliac catheter. Immediately after the bupivacaine dose, isoflurane was stopped and mechanical ventilation was continued with about 100% oxygen, with all animals receiving an infusion of either saline or about 30% by wt. solution of Intralipid® via the internal jugular catheter. In each case, the initial infusion rate was about 7.5ml/kg bolus over 30 seconds, followed

by a steady-state rate of about 3 ml/kg/min for about 2 minutes. Chest compressions were given during infusion for any animal experiencing more than about 15 seconds of asystole. Survival was scored about 5 minutes after the bupivacaine bolus and required both heart rate greater than about 100 beats per minute and systolic blood pressure greater than about 60 mmHg. Isoflurane at a concentration of about 1.75% was restarted whenever the blood pressure or heart rate met the survival criteria. Thus, a difference in survival between control and treated animals required rapid reversal of the cardiotoxic effects of a potentially fatal bupivacaine dose.

Preliminary experiments with this protocol established the bupivacaine bolus dose ranges necessary to achieve groups with 100% survival, 100% mortality and at least one intervening dose for both control and lipid treatment. These were 10 mg/kg, 12.5 mg/kg, and 15 mg/kg for the controls and 15 mg/kg, 17.5 mg/kg, 20 mg/kg and 22.5 mg/kg for lipid treated animals.

This resuscitation protocol provided a stringent test of efficacy of the lipid emulsion composition in treating bupivacaine induced cardiovascular collapse. The short fixed injection interval (10 seconds) modeled the clinical occurrence of a rapid intravascular bupivacaine injection. The experimental results showed about 48% increase in the bupivacaine LD_{50} when resuscitation included lipid infusion (from about 12.5 mg/kg to 18.5 mg/kg). At 15 mg/kg, a usually fatal bupivacaine dose, the lipid infused animals survived.

As illustrated by the foregoing results, lipid infusion reduces bupivacaine-associated cardiotoxicity. Partition experiments suggest that the primary benefit of lipid infusion results from a lipid sink effect where the poison is drawn from the blood into the non-aqueous component of the emulsion thereby reducing the amount of toxin in the cells such that toxicity is reversed. Other mechanisms may also be active. These observations suggest that the use of intravenously infused lipid emulsions can reverse toxic effects, particularly the cardiotoxic effects of lipophilic or ampiphilic agents. An important parameter in the design of such emulsion compositions is the partition coefficient of the toxic agent in the emulsion which can be readily determined by methods such as those described above.

Example 3

Emulsion Delivery Device

A delivery device 10 constructed in accordance with the teachings of the invention is illustrated in FIG. 2. As shown in that figure, the delivery device 10 includes a vented spike 12 for insertion into an intralipid (lipid emulsion)-containing bottle or other suitable container (not shown) suspended from an IV pole in a conventional manner. The vented spike 12 is in communication with a drip chamber 14 which is, in turn, in communication with a main delivery channel implemented by a length of tubing 16 which preferably has a large bore. The bottom end of the tubing 16 is provided with a connecting means, which preferably is a conventional luer lock 18.

To control the flow of fluid from the drip chamber 14, the delivery device 10 is provided with a conventional roller clamp 20 but other means for regulating fluid flow are also contemplated. The roller clamp 20 operatively engages the outer surface of the tubing 16 immediately below the drip chamber 14. By adjusting the roller clamp 18, health care personnel can regulate the rate at which fluid exits the drip chamber 14.

In accordance with one aspect of the invention, the delivery device 10 is provided with a length of extension tubing 22. As shown in Fig. 2, one end of the extension tubing 22 is in fluid communication with the large bore tubing 16. The extension tubing 22 may be integrally formed with or otherwise connected with the large bore tubing 16. The opposite end of the extension tube 22 is coupled to a syringe 24. The syringe 24 is preferably implemented with one or more finger rings 26 and a thumb ring 28 on its piston 30. Significantly, the syringe 24 can be suspended from the IV pole by its thumb ring 28. Suspending the syringe 24 in this manner prevents the extension tubing 22 and the syringe 24 from kinking when the injection fluid (emulsion) is just dripping and not being pushed from the syringe 24 via the piston 30.

For the purpose of controlling the direction of fluid flow through the device 10, the delivery device is provided with two one-way valves 32, 34. Preferably ball valves are utilized in the invention. As shown in FIG. 2, a first one of the ball valves 32 is located above the junction of the large bore tubing 16 and the extension

tube 22. This ball valve 32 prevents fluid flowing through the extension tube 22 from backing up the large bore tubing 16 towards the drip chamber 14. The second ball valve 32 is located below the junction of the large bore tubing 16 and the extension tube 22. This second ball valve 34 prevents fluid from passing back up the main delivery channel towards the syringe 24.

Preferably, the thumb ring 28 of the syringe 24 is suspended from the IV pole at substantially the same height as the top of the intralipid bottle. As a result, the length of the extension tubing 22 is preferably defined by the distance from the ball valve 32 to the IV pole. This distance is, in turn, defined by the length of the tubing 16 from the vented spike 12 to the ball valve 32 and the length of the intralipid bottle. Preferably, the length of the extension tubing 22 is selectively within these parameters to avoid any kinking when the syringe 24 is suspended from the IV pole.

While the above-described delivery device is particularly useful for administering the emulsions of the present invention, it is readily apparent that it may also be used to administer therapeutic or prophylactic substances.

The foregoing is intended to be illustrative of the invention but not limiting. Numerous variations and modifications of the present invention may be effected without departing from the spirit and scope of the invention.

All of the references cited herein are incorporated by reference in their entirety.

WE CLAIM:

1. A lipid emulsion composition which comprises an oil, an emulsifier, a tonicity modifier, and water.

2. The lipid emulsion composition of claim 1 further comprising a surfactant, a co-solvent, a bacteriostat, a preservative, an active ingredient, and an adsorbent.

3. The lipid emulsion composition of claim 1 wherein the oil is selected from the group consisting of monoglycerides, diglycerides, triglycerides, and mixtures thereof.

4. The lipid emulsion composition of claim 1 wherein the oil is a plant oil.

5. The lipid emulsion composition of claim 4 wherein the plant oil is selected from the group consisting of soybean oil, cotton seed oil, safflower oil, corn oil, coconut oil, sesame oil, peanut oil, olive oil and mixtures thereof.

6. The lipid emulsion composition of claims 1 or 2 wherein the oil is selected from the group consisting of soybean oil, fish oil, animal oil, mineral oil, and chemically-synthesized oil.

7. The lipid emulsion composition of claims 1, 2, 3, 4, or 5 wherein the emulsifier is a phospholipid.

8. The lipid emulsion of claim 6 wherein the emulsifier is a phospholipid.

9. The lipid emulsion composition of claims 1, 2, 3, 4, or 5 wherein the phospholipid is selected from the group consisting of egg yolk phospholipids, hydrogenated egg yolk phospholipids, soybean phospholipids, hydrogenated soybean phospholipids, and mixtures thereof.

10. The lipid emulsion composition of claim 6 wherein the phospholipid is selected from the group consisting of egg yolk phospholipids, hydrogenated egg yolk phospholipids, soybean phospholipids, hydrogenated soybean phospholipids, and mixtures thereof.

11. The lipid emulsion composition of claim 7 wherein the phospholipid is selected from the group consisting of egg yolk phospholipids, hydrogenated egg yolk phospholipids, soybean phospholipids, hydrogenated soybean phospholipids, and mixtures thereof.

12 The lipid emulsion composition of claim 8 wherein the phospholipid is selected from the group consisting of egg yolk phospholipids, hydrogenated egg yolk phospholipids, soybean phospholipids, hydrogenated soybean phospholipids, and mixtures thereof

13. The lipid emulsion composition of claims 1, 2, 3, 4, or 5 wherein the emulsifier is a lecithin.

14. The lipid emulsion composition of claim 6 wherein the emulsifier is a lecithin.

15. The lipid emulsion composition of claim 7 wherein the emulsifier is a lecithin.

16. The lipid emulsion composition of claim 8 wherein the emulsifier is a lecithin.

17. The lipid emulsion composition of claims 1, 2, 3, 4, or 5 wherein the tonicity modifier is selected from the group consisting of glycerin, sorbital, polyoxyethylated hydrocarbons, and C_6-C_{20} saturated or unsaturated aliphatic acids.

18. The lipid emulsion composition of claim 6 wherein the tonicity modifier is selected from the group consisting of glycerin, sorbital, polyoxyethylated hydrocarbons, and C_6-C_{20} saturated or unsaturated aliphatic acids.

19. The lipid emulsion composition of claim 7 wherein the tonicity modifier is selected from the group consisting of glycerin, sorbital, polyoxyethylated hydrocarbons, and C_6-C_{20} saturated or unsaturated aliphatic acids.

20. The lipid emulsion composition of claim 8 wherein the tonicity modifier is selected from the group consisting of glycerin, sorbital, polyoxyethylated hydrocarbons, and C_6-C_{20} saturated or unsaturated aliphatic acids.

21. The lipid emulsion composition of claim 1 wherein the tonicity modifier is glycerin.

22. The lipid emulsion composition of claim 1 wherein the oil is present in an amount in the range of about 10 to about 30 percent by weight based on the weight of the composition

23. The lipid emulsion composition of claim 1 wherein the surfactant is present in an amount in the range of about 1 to about 5 percent by weight based on the weight of the composition.

24. The lipid emulsion composition of claim 1 wherein the water is present in an amount in the range of about 70 to about 90 percent by weight based on the weight of the composition.

25. The lipid emulsion composition of claim 1 wherein the emulsion comprises particles in the range of about 0.25 microns to about 0.75 microns in diameter.

26. A lipid emulsion composition comprising about 20 weight percent soybean oil, about 2 weight percent glycerin, about 1 weight percent egg yolk phospholipid, and about 80 weight percent water.

27. A method for removing a toxin from the circulation which comprises infusing a lipid emulsion composition intravenously whereby the toxin permeates the lipid emulsion composition and is withdrawn from the bloodstream.

28. The method of claim 21 wherein the lipid emulsion composition comprises about 20 weight percent soybean oil, about 2 weight percent glycerin, about 1 weight percent egg yolk phospholipid, and about 80 weight percent water.

29. The method of claim 22 wherein the lipid emulsion composition is intravenously infused at an initial rate in the range of about 7.5 milliliters per kilogram per minute for a time period of about 30 seconds followed by a steady-state rate in the range of about 3 milliliters per kilogram per minute for a time period of about 2 minutes

30. A delivery device comprising:

a vented spike for insertion into a lipid-emulsion-containing container;

a drip chamber in communication with the vented spike;

a main delivery channel implemented by a length of tubing in communication with the drip chamber;

a connecting means contacting the bottom end of the large bore tubing;

a means for fluid rate regulation operatively engaged to the outer surface of the tubing immediately below the drip chamber;

a length of extension tubing in communication with the tubing;

a syringe coupled to the length of extension tubing opposite to the drip chamber;

a one-way valve joined to the large bore tubing above the length of extension tubing; and

a second one-way valve joined to the large bore tubing below the extension tubing.

31. The delivery device of claim 24 wherein the syringe is implemented with one or more finger rings and a thumb ring on its piston.

32. The delivery device of claim 25 wherein the syringe is suspended by the thumb ring from a pole.

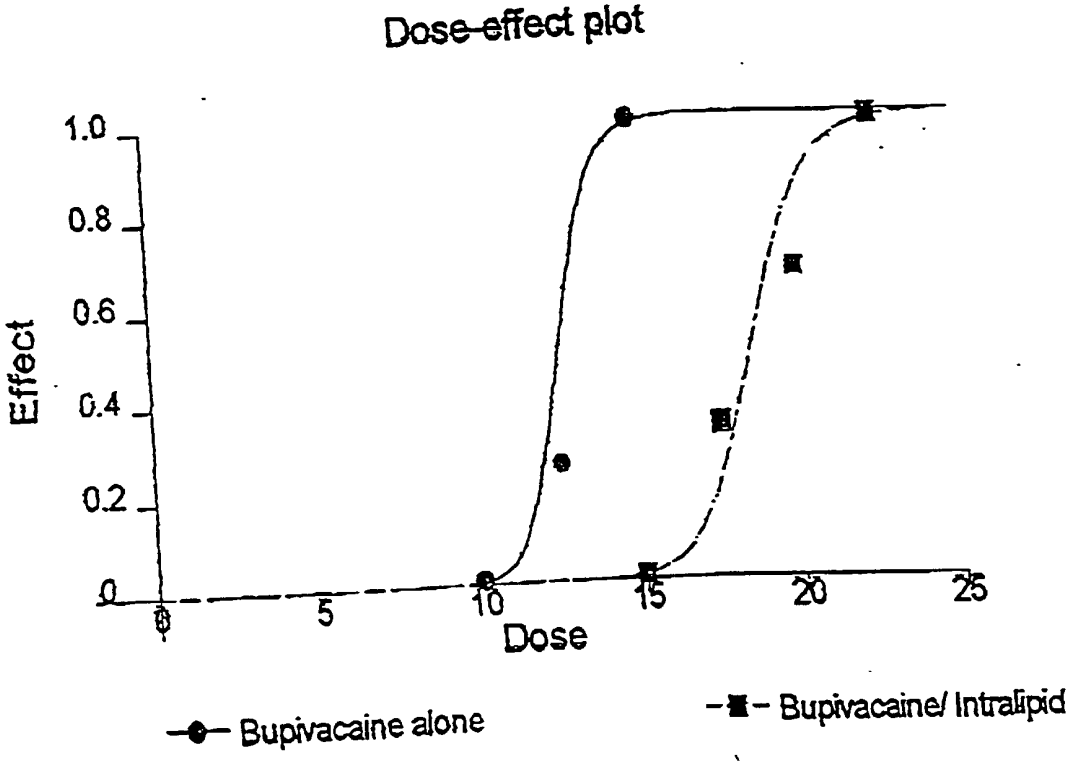
33. The delivery device of claim 26 wherein the thumb ring of the syringe is suspended at about the height of the top of the lipid-emulsion-containing container.

34. The delivery device of claim 27 wherein the length of extension tubing is about the length of the distance from the first valve to the pole.

35. The delivery device of claim 28 wherein the distance from the first valve to the pole is about the length from the vented spike to the first valve and the length of the lipid-emulsion-containing container.

ABSTRACT

Lipid emulsion compositions and methods of using such composition via intravenous infusion to reduce the bioavailability and toxicity of poisonous agents in the bloodstream.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10
15
20
25
Dose
Bupivacaine alone
Bupivacaine/ Intralipid

Figure 1

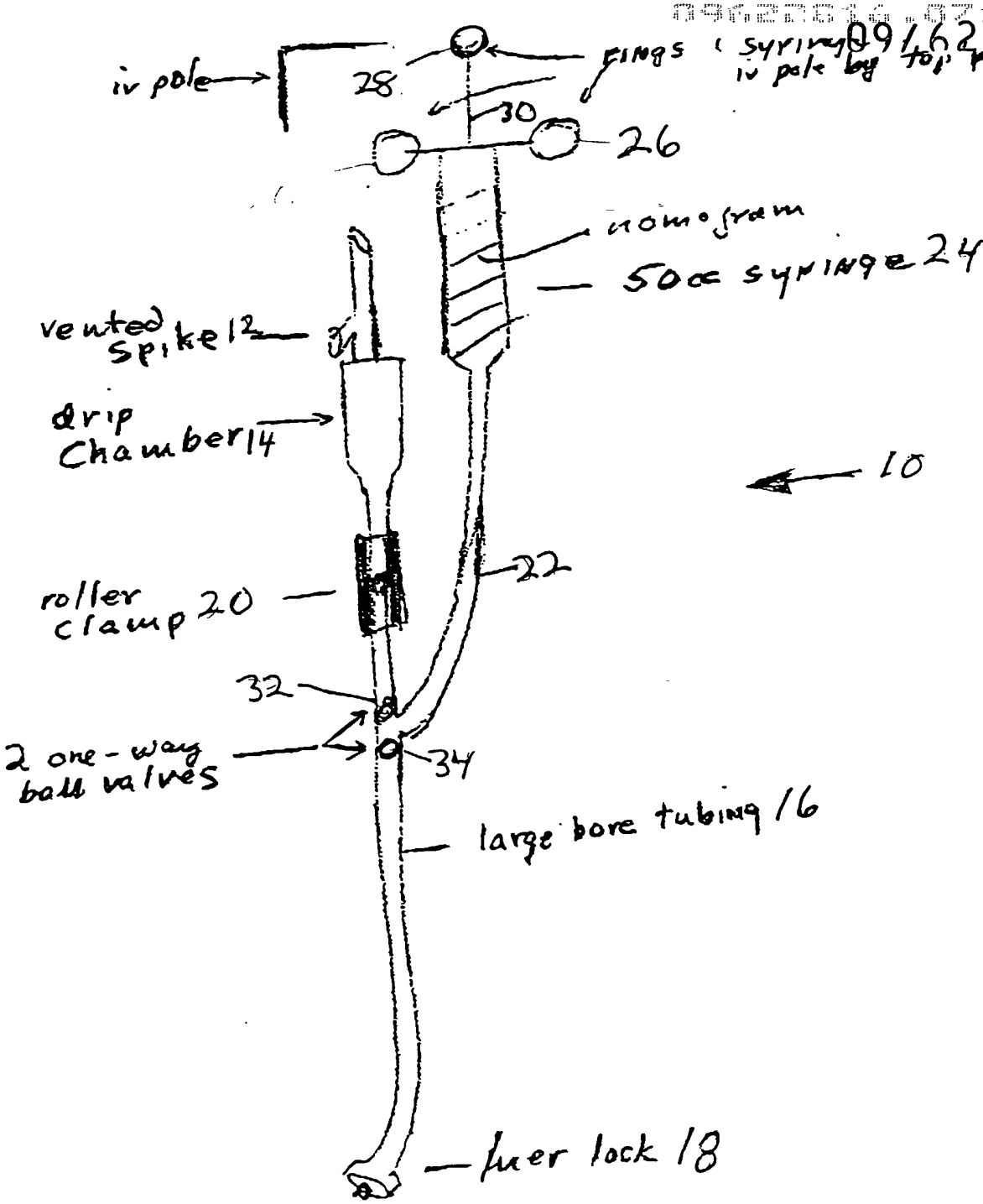

FIG. 2

DECLARATION FOR PATENT APPLICATION AND POWER OF ATTORNEY

As below named inventors, we hereby declare that our residences, post office addresses and citizenships are as stated below next to our respective names; we believe that we are the original and joint inventors of the subject matter which is claimed and for which a patent is sought on the invention entitled "**LIPID EMULSIONS IN THE TREATMENT OF SYSTEMIC POISONING,**" the specification of which was filed on **August 23, 2000,** as Application Serial No. **09/622,816,** which claims priority to International Application No. **PCT/US99/03805,** filed on **February 22, 1999.** We hereby state that we have reviewed and understand the contents of the above-identified specification, including the claims. We acknowledge the duty to disclose to the Patent and Trademark Office all information known to us to be material to patentability as defined in 37 C.F.R. §1.56.

We hereby claim foreign priority benefits under 35 U.S.C. §119 of any foreign application(s) for patent or inventor's. We certificate or of any PCT international application(s) designating at least one country other than the United States of America listed below and have also identified below any foreign application(s) for patent or inventor's certificate or any PCT international application(s) designating at least one country other than the United States of America filed by us on the same subject matter having a filing date before that of the application(s) of which priority is claimed:

			Priority Claimed	
PCT/US99/03805	International	22/02/1999	☒	☐
(Application Serial Number)	(Country)	(Day/Month/Year Filed)	Yes	No
			☐	☐
(Application Serial Number)	(Country)	(Day/Month/Year Filed)	Yes	No

We hereby claim the benefit under 35 U.S.C. §119(e) of any United States provisional application(s) listed below:

(Application Serial Number)	(Day/Month/Year Filed)
(Application Serial Number)	(Day/Month/Year Filed)

We hereby claim the benefit under 35 U.S.C. §120 of any United States application(s) or PCT international application(s) designating the United States of America listed below and, insofar as the subject matter of each of the claims of this application is not disclosed in the prior application(s) in the manner provided by the first paragraph of 35 U.S.C. §112, we acknowledge the duty to disclose to the Office all information known to us to be material to patentability as defined in 37 C.F.R. §1.56 which occurred between the filing date of the prior application(s) and the national or PCT international filing date of this application:

PCT/US99/03805	22/02/1999	
(Application Serial Number)	(Day/Month/Year Filed)	(Status-Patented, Pending or Abandoned)
(Application Serial Number)	(Day/Month/Year Filed)	(Status-Patented, Pending or Abandoned)

We hereby declare that all statements made herein of our own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under 18 U.S.C. §1001 and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

POWER OF ATTORNEY: we hereby appoint as my attorneys, with full powers of substitution and revocation, to prosecute this application and transact all business in the Patent and Trademark Office connected therewith:

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City (Zip)	City (Zip)
State or Country	State or Country
Date ☒	Signature ☒

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PATENT
Attorney's Docket No: 27611/34525A

Applicant or Patentee: Guy Weinberg and Paul Hertz

Serial or Patent No: To be assigned

Filed or Issued: Herewith

For: LIPID EMULSIONS IN THE TREATMENT OF SYSTEMIC POISONING

VERIFIED STATEMENT (DECLARATION) CLAIMING SMALL ENTITY STATUS (37 CFR 1.9(f) and 1.27(d)) -- NONPROFIT ORGANIZATION

I hereby declare that I am an official empowered to act on behalf of the nonprofit organization identified below:

NAME OF ORGANIZATION: The Board of Trustees of the University of Illinois

ADDRESS OF ORGANIZATION: 352 Henry Administration Building
506 South Wright Street, Urbana, Illinois 61801

TYPE OF ORGANIZATION

☒ UNIVERSITY OR OTHER INSTITUTION OF HIGHER EDUCATION

☐ TAX EXEMPT UNDER INTERNAL REVENUE SERVICE CODE (26 USC 501(a) and 501(c)(3))

☐ NONPROFIT SCIENTIFIC OR EDUCATIONAL UNDER STATUTE OF STATE OF THE UNITED STATES OF AMERICA

(NAME OF STATE____________________)
(CITATION OF STATUTE____________________)

☐ WOULD QUALIFY AS TAX EXEMPT UNDER INTERNAL REVENUE SERVICE CODE (26 USC 501 (a) and 501 (c)(3)) IF LOCATED IN THE UNITED STATES OF AMERICA

☐ WOULD QUALIFY AS NONPROFIT SCIENTIFIC OR EDUCATIONAL UNDER STATUTE OF STATE OF THE UNITED STATES OF AMERICA IF LOCATED IN THE UNITED STATES OF AMERICA
(NAME OF STATE ____________________)
(NAME OF STATUTE____________________)

I hereby declare that the nonprofit organization identified above qualifies as a nonprofit organization as defined in 37 CFR 1.9(e) for purposes of paying reduced fees under Section 41(a) and (b) of Title 35, United States Code with regard to the

invention entitled LIPID EMULSIONS IN THE TREATMENT OF SYSTEMIC POISONING by
inventor(s) Guy Weinberg and Paul Hertz described in

☒ The specification filed herewith.

☐ Application Serial No. _____, filed_____ .

☐ Patent No. _____ , issued _____.

I hereby declare that rights under contract or law have been conveyed to and remain with the nonprofit organization regarding the above-identified invention. If the rights held by the nonprofit organization are not exclusive, each individual, concern or organization having rights in the invention is listed below, and must file separate verified statements averring to their status as small entities and that no rights to the invention are held by any person, other than the inventor, who would not qualify as an independent inventor under 37 CFR 1.9(c), if that person made the invention, or by any concern that would not qualify as a small business concern under 37 CFR 1.9(d), or a nonprofit organization under 37 CFR 1.9(e).

FULL NAME: _____
ADDRESS: _____

☐ INDIVIDUAL ☐ SMALL BUSINESS CONCERN ☐ NONPROFIT ORGANIZATION

I acknowledge the duty to file, in this application or patent, notification of any change in status resulting in loss of entitlement to small entity status prior to paying, or at the time of paying, the earliest of the issue fee or any maintenance fee due after the date on which status as a small entity is no longer appropriate. (37 CFR 1.28(b)).

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application, any patent issuing thereon, or any patent to which this verified statement is directed.

NAME OF PERSON SIGNING: Craig S. Bazzani

TITLE IN ORGANIZATION: Comptroller and Vice President for Finance

ADDRESS OF PERSON SIGNING: 349 Henry Administration Building
506 S. Wright Street, Urbana, IL 61801

SIGNATURE: Craig S. Bazzani Date: August 22, 2000